

Draft Genome Sequence of *Methylobacterium buryatense* Strain 5G, a Haloalkaline-Tolerant Methanotrophic Bacterium

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Robust growth of the gammaproteobacterium *Methylobacterium buryatense* strain 5G on methane makes it an attractive system for CH₄-based biocatalysis. Here we present a draft genome sequence of the strain that will provide a valuable framework for metabolic engineering of the core pathways for the production of valuable chemicals from methane.

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Microbial utilization of methane is a key step in the carbon cycle (1–3). Methanotrophs provide an attractive platform for production of commodity chemicals and biofuels from natural gas/renewable biogas (4–6). Over the last 10 years, several novel methane-utilizing microbes have been isolated in pure culture but only a few, including *Methylobacterium buryatense* strain 5G, show robust growth on methane (5–7).

The draft genome was generated at the Department of Energy (DOE) Joint Genome Institute using Illumina sequencing (8). A short-insert paired-end library (insert size of 270 bp) generated 7.25 Mbp of data (<http://www.jgi.doe.gov/>). The initial draft data were assembled with Allpaths version 39750 and computationally shredded into 10-kbp overlapping fake reads (9). The initial data were also assembled with Velvet, version 1.1.05 (10), computationally shredded into 1.5-kbp overlapping fake reads, reassembled with Velvet, and shredded into 1.5-kbp overlapping fake reads. The fake reads from the Allpaths and two Velvet assemblies, as well as a subset of the Illumina CLIP paired-end reads, were assembled using parallel Phrap, version 4.24 (High Performance Software, LLC). Possible misassemblies were corrected by manual editing in Consed (11–13). Gap closure was accomplished using repeat resolution software and sequencing of bridging PCR fragments with Sanger and/or PacBio technologies (C. Han, W. Gu, unpublished). Fifty-three PCR PacBio consensus sequences were used to close gaps and to improve the quality of the final sequence.

The total estimated size of the genome is 5.4 Mb with an average coverage of 1,343×.

Comparative genome analysis of strain 5G and two other *Methylobacterium* species, *M. album* strain BG8 and *M. alcaliphilum* strain 20Z, revealed that 5G was most similar to *M. alcaliphilum*, sharing approximately 70% of its proteome at 90% protein sequence identity.

We identified genes encoding membrane-associated methane monooxygenase, soluble methane monooxygenase and an associated chaperon and a transcriptional activator (14), pyrroloquinoline quinone (PQQ)-dependent methanol dehydrogenase, an associated *c*-cytochrome, genes for enzyme assembly and PQQ biosynthesis, tetrahydromethanopterin- and tetrahydrofolate-linked C₁-transfer pathways, two formate dehydrogenases, and the ribulose monophosphate pathway. The Embden-Meyerhof-Parnas pathway, the Entner–Doudoroff pathway, and the pentose phosphate pathway (transaldolase variant) are predicted. As with the genomes of other gammaproteobacterial methanotrophs, the genome of *M. buryatense* 5G encodes all genes essential for operation of the citric acid cycle and the serine cycle, except for phosphoenolpyruvate carboxylase, isocitrate lyase, and the ethylmalonyl pathway (15,16).

Genes for urea uptake and hydrolysis, assimilatory nitrate/nitrite reduction, dissimilatory nitric oxide reduction, and ammonium uptake were identified. A gene homologous to hy-

droxylamine oxidoreductase is present (17, 18). The ammonium assimilation inventory includes genes for glutamate and alanine dehydrogenases, glutamate synthase/glutamine synthetase, serine-pyruvate/serine-glyoxylate, and aspartate aminotransferases (19). Genes essential for ectoine biosynthesis were identified.

Nucleotide sequence accession numbers. The *Methylobacterium buryatense* 5G genome sequence was deposited in GenBank/EMBL under the accession numbers [AOTL01000000](#) and [KB455575](#) and [KB455576](#).

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